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1: Biochim Biophys Acta. 1990 Apr 6;1048(2-3):297-302. Related Articles, Links

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Isolation and characterization of clones for the rat hepatic lipase gene upstream regulatory region.

Sensel MG, Legrand-Lorans A, Wang ME, Bensadoun A.

Division of Nutritional Sciences, Cornell University, Ithaca, NY 14853.

Genomic clones for 2287 nucleotides of the 5' flanking region, 135 nucleotides of the first exon, and 283 nucleotides of the first intron of the hepatic lipase gene were characterized. The predominant start site for transcription was identified by primer extension and S1 nuclease analyses to be 50 bases upstream of the ATG initiation codon. Based on the location of the major transcription start site, the functional TATA box is located 29 nucleotides upstream. Putative response elements for AP-2, cAMP, OCT-1, C/EBP, estrogen, glucocorticoids, sterols and thyroid hormone were located in this gene. Also a putative liver-specific element for apolipoproteins, C3P, was identified.

PMID: 2322583 [PubMed - indexed for MEDLINE]

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1: Gene. 1996 Nov 21;180(1-2):69-80. Related Articles, Links

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Transcription of the human hepatic lipase gene is modulated by multiple negative elements in HepG2 cells.

Oka K, Ishimura-Oka K, Chu MJ, Chan L.

Department of Cell Biology, Baylor College of Medicine, Houston, TX 77030, USA.

The expression of the hepatic lipase (HL) gene is highly tissue specific. In order to identify cis-acting elements which regulate the expression of this gene in the liver, multiple deletion mutants of the 5'-flanking region of the HL gene fused to the human growth hormone gene were transfected in HepG2 cells, which normally produce HL. Transient expression assays indicated the presence of negative (at nucleotides (nt) -1576(/)-1342 and -623(/)-407) and positive (at nt -1862(/)-1576 and -50(/)-9) regulatory elements. Transfection of HeLa cells, which do not produce HL, with the same deletion constructs resulted in a similar pattern of promoter activities. However, additional negative (nt -138/-50) and positive (nt -407(/)-138) elements were found. DNase I footprint analysis of the proximal and distal HLpromoter sequences with HepG2 and HeLa cell nuclear extracts identified seven protected regions: A, nt -1540(/)-1527; B, -1505(/)-1473; C, -1467(/)-1460; D, -592(/)-577; E, -565(/)-545; F, -234(/)-220; and G, -70(/) -48. Sites A, B, C, D and E were located within regions containing negative regulatory elements. In order to determine which nuclear factor interacts with the negative elements, sites B, D and E were mutated and the effects of mutation on competition in a gel retardation assay and on promoter activity were studied. When the binding motif for AP1 in sites B, D and E was mutated, the specific DNA-protein complexes were not competed with the mutant oligonucleotides and promoter activity increased twofold. The magnitude of the increase is less than expected from the deletion analysis, and simultaneous mutations did not cause further increase in promoter activity, which suggests that other sites are involved in this negative modulation. These results suggest that the transcription of the HLgene in HepG2 cells is negatively modulated by multiple cis-acting negative elements and AP1-like nuclear factor may play some role in this modulation.

PMID: 8973349 [PubMed - indexed for MEDLINE]

1: Biochemistry. 1993 Sep 21;32(37):9657-67. Related Articles, Links

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Identification of a cis-acting negative DNA element which modulates human hepatic triglyceride lipase gene expression.

Hadzopoulou-Cladaras M, Cardot P.

Department of Medicine, Boston University School of Medicine, Housman Medical Research Center, Massachusetts 02118.

The promoter fragment -1550/+129 of the human hepatic triglyceride lipase (HTGL) gene drives the expression of the CAT gene in HepG2 cells, albeit at very low levels. Transient transfections in HepG2 and HeLa cells of 5' deletion constructs indicated that the regulatory elements that control this expression are located in the proximal region of the gene. DNase I footprint analysis with DNA fragments spanning the region -483 to +129 and rat liver nuclear extracts identified eight protected regions, four upstream of the transcription initiation site (A, -28 to -75; B, -96 to -106; C, -118 to -158; D, -185 to -255) and four in the first exon of the gene (E1, -5 to +20; E2, +36 to +55; E3, +58 to +83; E4, +86 to +107). DNA binding and footprinting analysis demonstrated that the region -75 to -43 within footprint A binds to the liver-specific transcription factor HNF1. The region +28 to +129 contains a functional negative regulatory element (NRE) since deletion of this region results in a 17-fold increase in CAT activity. The NRE can act independent of orientation and position and repress transcription driven by heterologous promoters. DNA binding assays using native and fractionated liver nuclear extracts identified two transcription factors that bind to element E2 and also to element E3. A dinucleotide mutation in element E2 which causes derepression of the HTGL gene by 10-fold also abolishes the binding of these two activities. Transfection experiments showed that deletion of the NRE allows expression of reporter constructs in HeLa cells, indicating that the NRE may play a determinant role for the expression of HTGL gene in hepatic cells.

PMID: 8373770 [PubMed - indexed for MEDLINE]

1: Science. 1998 Jul 3;281(5373):61-3.

Going the distance: a current view of enhancer action.

Blackwood EM, Kadonaga JT.

Department of Biology and Center for Molecular Genetics, University of California, San Diego, La Jolla, CA 92093-347, USA.

In eukaryotes, transcription of genes by RNA polymerase II yields messenger RNA intermediates from which protein products are synthesized. Transcriptional enhancers are discrete DNA elements that contain specific sequence motifs with which DNA-binding proteins interact and transmit molecular signals to genes. Here, current models regarding the role of enhancers in the regulation of transcription by RNA polymerase II are presented.

Publication Types:
- Review
- Review, Tutorial

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